IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:

WANG

Application No.: 09/870,353

Filed: May 30, 2001

For: IMPROVED NUCLEIC ACID MODIFYING ENZYMES

Examiner: Richard Hutson

Technology Center/Art Unit: 1652

RULE 132 DECLARATION

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Sir:

I, Dr. Peter Vander Horn, being duly warned that willful false statements and the like are punishable by fine or imprisonment or both, under 18 U.S.C. § 1001, and may jeopardize the validity of the patent application or any patent issuing thereon, state and declare as follows:

1. All statements herein made of my own knowledge are true and statements made on information or belief are believed to be true. The Exhibits (1 - 10) attached hereto are incorporated herein by reference.

2. I received a Ph.D. in microbiology from Cornell University in 1991. A copy of my curriculum vitae is attached as **Exhibit 1**.

3. I am presently employed by MJ Bioworks, Inc. as Vice President of Research, Development, and Engineering. I am primarily responsible for supervising research teams working to improve our scientific instrumentation products. MJ Bioworks is the assignee of the subject patent application.

4. I have read and am familiar with the contents of the application. As I understand the bases for the outstanding rejections, the Examiner believes that the pending claims are overly broad and that it would take undue experimentation to identify members of the genus of non-specific double-stranded nucleic acid binding domains that are either recognized by polyclonal antibodies generated against Sso7d or have at least 50% identity to a 50 amino acid subsequence of Seq. ID No: 2 or a 75% identity to Sac7d.

5. The criteria set forth in the claims was intended to provide us with claim scope that embraced both naturally occurring proteins in the family of non-specific DNA binding Archaeal proteins as well as "Archaeal 7 kDa muteins". By Archaeal 7 kDa muteins, I am referring to man-made recombinantly produced proteins that are derived from naturally occurring proteins. In this context, muteins differ from their parent proteins by the introduction of amino acid changes where those changes do not markedly alter its **DNA binding** properties compared to the parent protein.

6. It is the intent of this declaration to explain in objective scientific reasons, why one of skill can identify working embodiments that fall within the scope of these claims with routine experimentation. In summary, there are three objective reasons and one subjective reason. The three objective reasons are: (i) that genetic variation or drift within the naturally occurring species of Archaeal 7 kDa proteins provides an initial road map for point mutations; (ii) that conventional knowledge of protein chemistry allows for us to predict that biological properties can be preserved so long as amino acid substitutions are conservative in their nature; and (iii) that knowledge of the three dimensional structure of these proteins when bound to DNA permits us to predict areas of non-criticality where substitutions may be freely introduced beyond mere conservative substitutions. As a subjective rationale, we must consider that the family of Archaeal 7 kDa proteins come from extremophilic bacteria that live in acidic environments above the melting temperature of DNA. This group of extremophiles includes many unexplored species that by virtue of their habitats are expected to have Archaeal 7 kDa-like DNA binding proteins. With so many species to be studied and so few cultured it is highly probable that additional members of the family will be discovered with even greater variation than those that are presently known and sequenced.

7. NATURAL VARIATION.

With regard to naturally occurring 7 kDa proteins in the family of Archaeal DNA-binding proteins, there are many family members reported in the literature. It is an accepted convention that proteins with E scores below 0.01 are unlikely to occur by chance and are therefore statistically related. Using Sso7d as a prototype, we studied the family of Archaeal DNA binding proteins reported in GenBank. We noted that there are at least 17 related members of the 7 kDa class of Archaeal proteins. The least related of which has an E value of $9x10^{-6}$.

The evolutionary relationship between the members of this family is made quite clear when you conduct a BlastP search comparing Sso7d to its family members. Using the default parameters provided by the specification on page 16, lines 7-11 with the "Low Complexity" filter set to off to permit us to align the entire 63 amino acids, we get the following results:

SEQ ID:2.	ATVKFKYKGEEKEVDISKIKKVWRVGKMISFTYDEGGGKTGRGAVSEKDAPKELLQMLEKQKK	Identity	Similarity
1) RNaseP3 of S:	atvkfkykgeekqvdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmmpetgkyfrhklpddypi	90%	95%
2) Sso7d	meismatvkfkykgeekevdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	100%	100%
3) Sso7d	matvkfkykgeekqvdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlakqkk	98%	100%
4) Sso7d	matvkfkykgeekevdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	100%	100%
5) Sso7d	atvkfkykgeekqvdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqk	98%	100%
6) Sso7d	matvkfkykgeekqvdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	98%	100%
7) Sso7d	atvkfkykgeekevdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	100%	100%
8) Sso7d	atvkfkykgeekqvdiskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	98%	100%
9) Ssh7B	mvtvkfkykgeekevdtskikkvwrvgkmisftydegggktgrgavsekdapkellqmlekqkk	98%	98%
10) Sso7d mutant	atvkfkykgeekqvdiskikkvwrvgkmisatydegggktgrgavsekdapkellqmlekqk	96%	98%
11) Sso7e/Sto7e	mvtvkfkykgeekevdiskikkvwrvgkmisftydd-ngktgrgavsekdapkellqmleksgkk	91%	93%
12) Sac7a	vkvkfkykgeekevdtskikkvwrvgkmvsftydd-ngktgrgavsekdapkelldmlarae	86%	91%
13) Sac7a/b/d	mvkvkfkykgeekevdtskikkvwrvgkmvsftydd-ngktgrgavsekdapkelldmlaraerekk	81%	90%
14) Sac7e	makvrfkykgeekevdtskikkvwrvgkmvsftydd-ngktgrgavsekdapkelmdmlaraekkk	79%	88%
15) 1SAP/Sac7	kvkfkykgeekevdtskikkvwrvgkmvsftydd-ngktgrgavsekdapkelldmlaraerekk	86%	91%
16) Sac7e	akvrfkykgeekevdtskikkvwrvgkmvsftydd-ngktgrgavsekdapkelmdmlaraekkk	79%	88%
17) Sso Dna binding protein	tvkfkykgeekqvdiskikkvxrvgkmisftydegxgk	92%	94%

From the above BLASTP data, we can see that the natural variation within the family extends to below 80% identity. At a minimum, it was the applicants' intent to encompass in a single claim all naturally occurring known variants of the DNA binding Archaeal protein family. But our knowledge of variants can be extended to include muteins by applying our knowledge of protein chemistry - knowledge that is both routine and predictable in its application.

8. MUTEINS CREATED BY COMBINING NATURALLY OCCURRING VARIATION.

Muteins of Archaeal 7 kDa proteins can be readily created by those of skill exploiting variation within the natural members of the family to create novel combinations of variations. In essence, the naturally occurring members are a road map to defining the critical amino acids from the non-critical amino acids.

A cursory review of the family reveals that the amino and carboxyl termini are not critical to the functionality of these proteins. The amino and carboxyl ends are very tolerant of substitutions and additions. They are sites of divergence between the homologues and the invention. As evidence of the robust nature of these proteins, we placed entire polymerase domains on both the carboxyl and the amino ends without interfering with binding. This was Dr. Wang's rationale for claiming sequence similarity to a 50-amino acid subsequence, rather than to the entire protein. Biological functionality appears to be determined by the conserved amino acids that form the internal core of these proteins (see Choli et al. (1988) *Biochimica et Biophysica Acta*, 950:193-203 at 202) (**Exhibit 2**). But even there the identity is not 100%.

9. MUTEINS CREATED BY INTRODUCTION OF CONSERVED SUBSTITUTIONS.

In addition to the introducing combinations of naturally occurring variations into a prototype 7 kDa binding protein, those of skill can also substitute *conserved* amino acids for naturally occurring ones that have not been found to vary in nature. Classic examples of such pairings are lysine and arginine, alanine and glycine, glutamine and asparagine, and aspartic acid and glutamic acid. All of which appear in this family of proteins. For example, there are 12 residues of Sso7d 63 residues in which natural variations are known. By substituting conserved amino acids for another 20 residues, we can easily produce a non-specific 7 kDa Archaeal mutein that would almost certainly work to improve processivity of a polymerase.

10. MUTEINS DERIVED FROM STUDIES OF THREE DIMENSIONAL ANALYSES.

We need not limit our muteins to combinations of naturally occurring amino acid variations nor to those that are unnatural but between amino acids of similar chemical properties. This is because the three dimensional structure of these proteins when interacting with DNA is known. See **Exhibit 3** Gao *et al.*

Knowledge of three dimensional features provides yet another strategy permitting protein chemists to engineer away from the native sequences because it provides structural activity relationships between the protein domains and DNA. Knowing which domains play a role in DNA binding and which are non-critical for binding permits us to think beyond mere conservative amino acid substitution and to allow for Archaeal 7 kDa muteins with lower percent identities than if we confined our mutein development strategy to the first two objective approaches.

Attached to this declaration as **Exhibits 4-8** are enlargements of figures derived from the data of Gao, et al. with an accession number of 1BNZ.[1] **Exhibit 4** is a ribbon diagram of the

[1] These figures are derived from the protein crystal coordinates that Gao submitted to the protein structure database. Submission is a requirement

crystal structure of Sso7d bound to DNA. The beta sheets of the protein are in yellow, the alpha helix is in green. Unstructured regions are in blue.

As predicted, the unstructured regions are sites where divergences from Sso7d among the group of related proteins cluster. One skilled in the art could place additional insertions into these sites that will decrease sequence identity in blast analyses. For example, a thermostable loop can be placed in the G37, G38, G39 turn.

In addition, the entire alpha helix (green) is highly mutable. This is evidenced by the fact that a great deal of natural variation of the homologs is observed in this domain. It should be noted that the naturally occurring mutations in this domain do appear to preserve the presence of an alpha helix and this region does not interact with the DNA substrate. Therefore, additional mutations could be introduced into the alpha helix (as long as they preserve the secondary structure) and serve to further lower the amino sequence identity compared to SEQ ID 2.

Using the three dimensional figures, those of skill could also take note that the differences in composition and length between Sso7 and Sac7 proteins cluster in the turns between beta sheets and in amino acids facing away from the DNA binding domain in the crystal structure. So these domains are also areas of plasticity.

The papers cited in the patent application describe several exposed lysine residues that are methylated *in vivo*. These sites are not involved in DNA binding but appear to be regulatory. As our work is independent of bacterial gene regulation, these lysines could be mutated so long as they do not interact with the DNA substrate. As can be seen in **Exhibits 5** though **8**, many of these lysine residues project away from the domain and do not interact with DNA. These residues are excellent candidates for mutagenesis. One skilled in the art would recognize that these could be changed to arginine residues without affecting DNA binding.

I was able to find 10 such sites by examining the crystal structure. **Exhibit 5** shows lysines 19, 40, 49, and 53 projecting away from the DNA binding surface of the protein. **Exhibit 6** also shows lysines 49, 61, and 64. Exhibit 7 shows lysine 63 and Exhibit 8 shows lysines 5 and 13. K to R derivatives already exist for positions 5 and 61, validating this approach. No divergence from the Sso7d sequence has been observed for the remaining 8 lysines, probably because of the regulatory role alluded to earlier. Mutating these lysines can yield an additional 8 differences from SEQ ID No. 2, or 13%.

---

similar to the requirement that sequences be deposited into Genbank with an accession number. The accession code for Sso7d protein bound to DNA is 1BNZ. The coordinates are viewed and turned into these figures using the program Cn3d, which is freely available at http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?db=Structure.

For these varied but objective reasons, one skilled in the art could with a combination of conserved substitutions, insertions, deletions, and exchanges of mutable sites construct DNA binding proteins that are very divergent from SEQ ID: 2 and Sac7d. I will discuss specific percentages later in this Declaration.

11. OTHER EXTREMOPHILES WILL HAVE ARCHAEAL 7 kDa LIKE PROTEINS.

Beyond the objective reasons presented above, there is a subjective reason why a percentage below 90% is needed to avoid routine engineering around the presently issued claims. As of today there have been many Archaeal 7 kDa proteins that have already been reported, it should be noted that these proteins are very abundant in *Sulfolobus* species. In fact, they are probably abundant in any organism that has to live in acid at >70°C chemolithotrophically. Here are *S. Solfataricus's* relatives many of which are expected to contain Sso7d-related proteins.

```
Archaea; Crenarchaeota; Thermoprotei; Sulfolobales
Sulfolobaceae
     Acidianus
          Acidianus ambivalens
          Acidianus brierleyi
          Acidianus infernus
          Acidianus tengchongenses
          Metallosphaera
          Metallosphaera prunae
          Metallosphaera sedula
          Metallosphaera sp. GIB11/00
          Metallosphaera sp. J1
          Metallosphaera sp. TA-2
          environmental samples
               uncultured Metallosphaera sp.

     Stygiolobus
          Stygiolobus azoricus
          environmental samples
               uncultured Stygiolobus sp.

     Sulfolobus
          Sulfolobus acidocaldarius
          Sulfolobus islandicus
          Sulfolobus metallicus
          Sulfolobus shibatae
          Sulfolobus solfataricus
          Sulfolobus thuringiensis
          Sulfolobus tokodaii
          Sulfolobus yangmingensis
          Sulfolobus sp.
          Sulfolobus sp. AMP12/99
          Sulfolobus sp. CH7/99
          Sulfolobus sp. FF5/00
          Sulfolobus sp. MV2/99
          Sulfolobus sp. MVSoil3/SC2
```

```
        Sulfolobus sp. MVSoil6/SC1
        Sulfolobus sp. NGB23/00
        Sulfolobus sp. NGB6/00
        Sulfolobus sp. NL8/00
        Sulfolobus sp. NOB8H2
        Sulfolobus sp. RC3
        Sulfolobus sp. RC6/00
        Sulfolobus sp. RCSC1/01
        Sulfolobus sp. RT8-4
        environmental samples
             uncultured Sulfolobus sp.

    Sulfurisphaera
        Sulfurisphaera ohwakuensis
```

So far only *Sulfolobus solfataricus* and *Sulfolobus tokodaii* genomes have been sequenced.

Given the range of divergence in Archaeal 7 kDa DNA binding proteins set forth above from a tiny portion of species sequenced, it will be trivial to find additional species of these DNA binding proteins that will have 70% or less homology to the presently known prototypes.

12. THE 90% LIMITATION OF THE '424 PATENT INVITES THOSE OF SKILL TO ENGINEER AROUND THE CLAIMS WITH EASE.

Let's look more specifically at the information that was available prior to filing the subject application. Dr. Wang's earlier patent US Pat. No. 6,627,424 ['424] issued with claims covering 90% identity to Sso7d and identity to Sac7d. Below I have created a paired table comparing the relative homology between Sso7d and Sac7d and Sac7d and Sac7e.

As you can see, close relatives of Sso7d, (i.e., Sac7a,b,d and e) are not covered by the recited percentage in our '424 patent claims. But a pair-wise alignment of these sequences to the two specific examples gives one a clear road map to implementing the invention with any of the naturally occurring homologues.

```
Sso7d alignment to Sac7d.
Sso7d: 1  MATVKFKYKGEEKEVDISKIKKVWRVGKMISFTYDEGGGKTGRGAVSEKDAPKELLQML---EKQKK 64  Identity Similarity
          M--VKFKYKGEEKEVD-SKIKKVWRVGKM+SFTYD+--GKTGRGAVSEKDAPKELL-ML---E++KK      80%      85%
Sac7d: 1  MVKVKFKYKGEEKEVDTSKIKKVWRVGKMVSFTYDD-NGKTGRGAVSEKDAPKELLDMLARAEREKK 66
```

Note: the percent identity changes to 82% and the similarity changes to 88% if Seq ID 2 is used. This is because Seq ID 2 is Sso7d without the MET. One skilled in the art would study the entire sequence.

```
Sac7d aligned to Sac7e (not covered in the '424 patent because it is 79% identical to Seq ID 2).
Sac7d: 1  MVKVKFKYKGEEKEVDTSKIKKVWRVGKMVSFTYDDNGKTGRGAVSEKDAPKELLDMLARAEREK 65  Identity Similarity
          M KV+FKYKGEEKEVDTSKIKKVWRVGKMVSFTYDDNGKTGRGAVSEKDAPKEL+DMLARAE++K       92%      98%
Sac7e: 1  MAKVRFKYKGEEKEVDTSKIKKVWRVGKMVSFTYDDNGKTGRGAVSEKDAPKELMDMLARAEKKK 65
```

Note: A 49 amino acid core sequence is completely identical.

Finding alternative species not covered by the allowed claims of the '424 patent, whether the above recited naturally occurring species or man-made muteins are trivial exercises for one skilled in the art. No reasonable protein chemist looking at this data would doubt that Sac7e could increase the processivity of polymerases if traded out for Sac7d in the constructs in Seq ID No. 9 and SEQ No. ID 10 of the '424 patent.

It is also helpful to take note that three of the references Dr. Wang cited in the patent (Choli et. al. **Exhibit 2,** Baumann et. al. **Exhibit 9**, and McAfee et. al. **Exhibit 10)** contain figures with sequence alignments of Sso7d homologues including Sac7d, Sac7a, and Sac7e. They are repeatedly described as structurally and functionally closely related proteins. The Sac7d construct (figure 2 of the application) was made to support that contention that these homologues would work. Dr. Wang clearly knew about and taught these proteins would work in the invention. No one skilled in the art that reads the patent specification and the referenced papers would have objective reasons to think it wouldn't work.

For these reasons, I submit that a 79% identity to Sso7d using naturally occurring variants is clearly enabled by the specification.

13. ROUTINELY INTRODUCING NON-NATURAL VARIATIONS LOWERS THE PERCENTAGE BELOW 79%.

Using natural variants as a road map a 79% identity is readily available. But man-made modifications can take this 79% identity lower. One can go lower in percent identity by merely combining known deviations from Sso7d. Using the family of Sac7 proteins as a road map one obtains the following hybrid sequence:

Hypothetical 7d: MVKVKVRFKYKGEEKQVDTSKIKKVGRVGKMVSATYDDNGKTGRGAVSEKDAPKELLDMLARAEREKK[2]

The hypothetical protein 7d is 76% identical to Sso7d as shown in the alignment below.

```
Sso7d:    ATVKFKYKGEEKEVDISKIKKVWRVGKMISFTYDEGGGKTGRGAVSEKDAPKELLQML---EKQKK 64
          --V+FKYKGEEK+VD-SKIKKV-RVGKM+SFTYD+--GKTGRGAVSEKDAPKELL-ML---E++KK
Hypothet: VKVRFKYKGEEKQVDTSKIKKVGRVGKMVSFTYDD-NGKTGRGAVSEKDAPKELLDMLARAEREKK 65
```

14. COMBINING ALL THE INFORMATION WILL LEAD ONE OF SKILL TO MUTEINS HAVING LESS THAN 60% SEQUENCE IDENTITY TO SAC7d.

Combining all of these changes together one can get a functional derivative of SEQ ID No. 2 with less than 60% amino acid identity in a blast search. One example of such a protein sequence is below.

---

[2] One known Sso7d divergence was not included in this alignment. The F34A mutation was not included because it is known to destabilize the protein. All other divergences are from functional proteins.

VKVRVRFKYK GEERQVDTSR IRKVGRVGKM VSATYDDACA AACNGRTGRG AVSERDAPRE LLDMLARAER ERR

We have identified other muteins of Sso7d that enhance polymerase performance. For instance: I^{21} to V, T^{47} to N, D^{56} to Y, and M^{64} to K in the above sequence. With one exception (I^{21} to V), these are not conserved changes; but, they are changes that do not affect core structures.

When all this information is combined, it would be straightforward to identify muteins with less than 60% identity to Sso7d that would still enhance polymerase performance.

15. THE ARCHAEAL 7 kDa PROTEINS ARE AN ANCIENT PROTEIN AND EXISTING EVOLUTIONARY DRIFT ESTABLISHES THE HIGH PROBABILITY THAT MUTEINS WITH 50% IDENTITY TO ANY KNOWN SPECIES CAN BE CREATED.

From an evolutionary perspective, this family of thermal stable DNA binding proteins is apparently quite ancient. There is a restriction endonuclease from *Methanococcus jannashii* (Results below)--another archaeon-- that a blast search of the Swissprot Database with Seq ID No. 2 will identify. The 47% identity of this DNA binding protein to Sso7d indicates that the DNA binding domain has been around for a long time and that with routine sequencing of genomes from the Archaeal family there will be many easily obtainable proteins with even less than 50% identity to Seq ID No. 2 that will work in the invention.

>gi|10954528|ref|NP_044167.1| M. jannaschii predicted coding region MJECL41 [Methanococcus jannaschii]

gi|12229988|sp|Q60296|T1SH_METJA Putative type I restriction enzyme MjaXP specificity protein (S protein) (S.MjaXP)

gi|2129054|pir||H64514 hypothetical protein MJECL41 - Methanococcus jannaschii plasmid pURB800

gi|1522674|gb|AAC37110.1| M. jannaschii predicted coding region MJECL41 [Methanococcus jannaschii]
Length = 432

Score = 30.0 bits (66), Expect = 8.5
Identities = 19/45 (42%), Positives = 24/45 (53%), Gaps = 1/45 (2%)

```
Query: 3  VKFKYKGEEKEVDISKIKKVWRVGKMISFTYDEGGGKTGRGAVSE 47
          VKF+++.E.KE.DI.KI.K.W.V.K.I.....+.....GG.T.....+.E
Sbjct: 5  VKFRWETEFKETDIGKIPKDWDV-KKIKDIGEVAGGSTPSTKIKE 48
```

Having provided multiple objective roadmaps to the creation of muteins, it needs to be said that actual function is always subject to empirical determination. To determine if the 7 kDa Archaeal muteins function as desired, the Examiner is asked to take note of the generic assay for DNA binding described on pages 18-19 of the specification. Here, the inventors present a generic method for readily and conveniently testing for operable species.

Based on the objective reasons set forth above, I submit that the creation of Archaeal 7 kDa muteins having 60 to 50% identity to native Archaeal 7 kDa is a matter of routine experimentation.

16. DEFINING THE PROTEINS BY THEIR ABILITY TO BIND TO ANTIBODIES GENERATED AGAINST A PROTOTYPE LIMITS THE PRIMARY AMINO ACID TO DEFINED STRUCTURE.

In addition to defining the invention by a percent identity, an alternative scope of claim protection was presented where the DNA binding proteins were defined as those recognized by polyclonal antibodies generated against specific Archaeal 7 kDa DNA binding proteins. The Examiner has rejected claims directed to non-specific double-stranded nucleic acid binding domains that are recognized by polyclonal antibodies generated against Sso7d. As I understand the rejection, the Examiner believes that the scope of this claim encompasses too many non-operable species to be considered allowable.

In the first instance, I would like to point out that the scope of proteins encompassed by the language is more limited than the claims where the proteins have 50% identity.

The use of immuno-crossreactivity to define proteins as related or unrelated is an old and well-recognized art. The specification, at pages 16-18 provides a routine and conventional means to compare unknown proteins with known proteins.

In addition, it is well-known in the art to use antisera as identification reagents to clone genes, based on the expression of a protein mediated by an expression vector. If the library source is one of the naturally-occurring relatives of *Sulfolobus sulfataricus* listed above, the probability that any cross-reacting gene obtained from the library would function to increase the processivity of polymerases is very high.

But naturally occurring proteins are not the only proteins that would be expected to cross react with polyclonal antisera generated against the prototype Archaeal 7 kDa proteins. One could easily envision muteins that would retain immuno-crossreactivity. To the extent that some may lack function; those inoperable embodiments could be rapidly distinguished from operable species using the prescribed assay set forth in the specification.

When these teachings are coupled with the generic assay for testing functionality of the proteins to non-specifically bind to DNA (see the specification at pages 18 and 19), I submit that there is no objective reason to doubt that the identification of many operable species with 50% or greater sequence identity with SSo7d or Sac7d with polyclonal antibodies specific to the two prototypes would be anything other than routine and expected.

This Declarant has nothing further to say.

Peter B. Vander Horn

Dated: 3/1/04

attachments: Exhibit 1-10

TOWNSEND and TOWNSEND and CREW LLP
Two Embarcadero Center, 8th Floor
San Francisco, California 94111-3834
Tel: 415-576-0200
Fax: 415-576-0300
KAW:jhd

60140599 v2